ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Jan. 1977, p. 132-141

Vol. 11, No. 1
Printed in U.S.A.

# Pharmacokinetics of Amoxicillin: Dose Dependence After Intravenous, Oral, and Intramuscular Administration

DANIEL A. SPYKER,* RAYMOND J. RUGLOSKI, ROBERT L. VANN, AND WILLIAM M. O'BRIEN

*Department of Internal Medicine, University of Virginia School of Medicine, Charlottesville, Virginia 22901,* and Beecham Laboratories, Bristol, Tennessee 37620*

Received for publication 16 September 1976

Amoxicillin was studied in normal subjects after intravenous, oral, and intramuscular administration of 250-, 500-, and 1,000-mg doses. Serum drug levels were analyzed using a two-compartment open model, as well as area under the curve (AUC) and urinary recovery. The variations of these pharmacokinetic parameters were then examined using the three-way analysis of variance and linear regression equations. These results confirmed nearly complete oral absorption: AUC was 93% of intravenous absorption, and urinary recovery was 86%. The intramuscular administration of amoxicillin results in complete and reliable absorption with peak drug levels, AUCs, and urinary recovery equivalent to oral dosage. The absorption of lyophilized amoxicillin after intramuscular injection resulted in an AUC that was 92% of intravenous absorption and urinary recovery of 91%. The peak serum levels, time to peak, and other pharmacokinetic parameters for intramuscular injection were nearly identical to those for oral administration. Kinetics of both intramuscular and oral administration exhibited dose-dependent absorption (absorption rate constant, 1.3/h for 250 mg and 0.7/h for 1,000 mg). This resulted in relatively later and lower peak serum levels for increasing dose. Total absorption, however, showed no dose dependence, as indicated by urinary recovery and AUC, which changed by less than 10%.

Amoxicillin (alpha-amino-*p*-hydroxybenzyl penicillin) is a new semisynthetic penicillin, similar in structure and spectrum of activity to ampicillin (14, 29). Amoxicillin produces higher serum levels than ampicillin with similar absorption and excretion kinetics (10, 21). We report a pharmacokinetic comparison of serum levels and urinary excretion data for amoxicillin administered by mouth (p.o.), intramuscularly (i.m.), and intravenously (i.v.), for three doses in in normal volunteers.

Abbreviations used in this report include the following: ANOVA, analysis of variance; RMS, root mean square; SEM, standard error of the mean; AUC, area under the curve (hours × micrograms per milliliter); alpha, slope of log concentration-early (distribution) phase, per hour; beta, slope of log concentration-late (elimination) phase per hour; $V_1$, volume of central (plasma) compartment, liters per kilogram; $V_2$, volume of peripheral (tissue) compartment, liters per kilogram; $V_d$, total volume of distribution (central+tissue), liters per kilogram; $K_{12}$, transfer rate constant (central to peripheral), per hour; $K_{21}$, transfer rate constant (peripheral to central), per hour; $K_a$, absorption rate constant, per hour; $K_e$, elimination rate constant, per hour; Do, dose of drug (milligrams); f, fraction of the drug absorbed.

A group of eight subjects was given amoxicillin i.v. in doses of 250, 500, and 1,000 mg and then subsequently received the same doses p.o. In this group, distribution and excretion were characterized using a two-compartment linear model. A second group of 24 subjects, separate from the above group, was given 250-, 500-, and 1,000-mg doses p.o. and also given the same doses via i.m. injection. The administration of three doses permitted the evaluation of a dose effect on drug absorption, distribution, and excretion.

## MATERIALS AND METHODS

**Description of subjects.** Subjects were healthy adult male volunteers ranging in age from 18 to 32, and in weight from 57 to 98 kg. They were selected on the basis of a complete medical history to exclude those having a history of hematological, hepatic, or renal disease, and those with a history of allergic sensitivity to penicillins. A complete blood count, urinalysis, and blood chemistry screen (including levels of total protein, albumin, calcium, inorganic phosphorus, cholestrol, glucose, blood urea nitrogen, uric acid, total bilirubin, alkaline phosphatase, serum glutamic oxalacetic transaminase, serum glutamic pyruvic transaminase, and lactic dehydrogenase) were performed on all subjects before start-

ing the study and repeated at the end of each day on which the drug was administered. Previously screened alternate subjects replaced the few who dropped out during the study.

**Administration of amoxicillin.** (Drugs were provided by Beecham Laboratories, Bristol, Tenn.)

Eight subjects participated in each of three crossovers, i.v. versus p.o., studies at dosages of 250, 500, and 1,000 mg of sodium amoxicillin. A 1-week washout period was observed between each dosage change, necessitating nearly 6 weeks of involvement. Half the subjects randomly received amoxicillin i.v., whereas the other half received the same dose of the drug p.o.; after a week interval, the routes of administration were crossed over. This was sequentially done through 250-, 500-, and 1,000-mg dosages, so that each subject received all three doses both i.v. and p.o. When administered i.v., amoxicillin was given as a 10-s bolus, and blood samples were drawn beforehand (at zero time) and at 1, 2, 5, 10, 15, 20, 25, 30, 45, 60, 75, and 90 min and 2, 2.5, 3, 4, and 6 h after dosing. From those receiving amoxicillin p.o., blood was drawn at zero time, and at 20, 40, 60, and 90 min and 2, 4, and 6 h after ingestion. Urine was collected over intervals of 0 to 1, 1 to 2, 2 to 3, 3 to 4, and 4 to 6 h.

In the i.m. versus p.o. study 24 volunteers participated. The same general procedures and crossover design as the i.v. versus p.o. study were followed, i.e., each subject received all three doses both i.m. and p.o. The dosages of amoxicillin were 250, 500, and 1,000 mg. (Lyophilized sodium amoxicillin was provided for i.m. injection.) Blood samples were taken immediately before (at zero time) and at 20, 40, 60, and 90 min and at 2, 4, 6, and 8 h after dosing. Urine was collected over intervals of 0 to 4 and 4 to 8 h.

**Bioassay.** Blood was collected in evacuated glass tubes without anticoagulant, and serum was obtained by centrifugation. All samples were stored in duplicate and deep frozen until assayed. The amoxicillin content of the serum and urine samples was determined by a large plate (13 by 16 inches [33 by 40 cm]) cup assay method using *Bacillus subtilis* (ATCC 6633) as the assay microorganism. As described elsewhere (3), the sensitivity of this assay is 0.5 $\mu$g/ml, and the precision is $\pm 5\%$. Complete standard curves were prepared for each assay using human serum or urine as appropriate; five samples were assayed per each plate (3).

**Data handling.** Data for each subject were entered via computer terminal and stored directly on disk files. Verification of these master data files was achieved by having a second person enter the data and the computer check for discrepancies. Thereafter, data were manipulated only by pharmacokinetic and statistical programs. Appropriate data files were constructed for each group of subjects at various stages in the analysis as described below. All programs were generalized (as to number of subjects, number of observations, number of parameters, etc.), and subsequent analysis required a minimum number of programs and virtually no operator intervention.

**Nonparametric analysis.** Nonparametric (model-independent) evaluation of the kinetic data was completed for each subject as an initial step. This included a regression analysis of log of concentration versus time for the following: (i) elimination rate constant from last three data points; (ii) distribution rate constant by exponential peeling (i.v. data); (iii) peak and time to peak from quadratic function fit to three highest drug levels (i.m. or p.o. data); and (iv) absorption delay and rate constant, derived using the method of Wagner and Nelson (30) (i.m. or p.o. data).

AUC was computed using the trapezoid approximation. Correction for the area after the last blood sample was based on the elimination rate constant [as mentioned in (i) above]. An individual plot for each administration trial, along with a summary of derived data, was plotted for each subject (Fig. 1 and Table 1).

**Two-compartment linear model.** The data were then examined using a two-compartment linear model that has been shown to adequately describe the course of many drugs in humans (24). All absorption and elimination takes place through the central compartment and thus, the absorption rate constant ($K_a$) and elimination rate constant ($K_e$) of the drug are characterized as processes involving the central compartment. The parameters $K_{12}$ and $K_{21}$ represent the rate constants of drug transfer between the central and peripheral compartments as summarized in Fig. 2.

General solutions for the two-compartment model have been detailed by Bischoff and Dedrick (4) and recently presented in the clinical context (11, 12). The solution for drug level in the central compartment ($C_1$) can be written as an exponential equation (Fig. 3).

Experience has shown that the absorption of tablets administered p.o. can usually be well described as a simple first-order, i.e., exponential, process with a time delay (18). The solution equation for central compartment concentration then becomes as shown in Fig. 4.

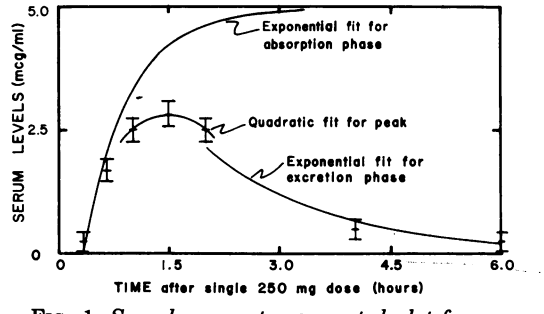


FIG. 1. *Sample computer-generated plot for nonparametric analysis. In this step, drug level data is examined without model assumptions. The best (in a least-squares sense) exponentials are fit for absorption and excretion, a quadratic is fit to the middle data to localize peak and time to peak, and area is computed using the trapezoid rule. Bars (‡) represent observed data. Accompanying printout is contained in Table 1.*

TABLE 1. *Sample of nonparametric analysis. Computer printout of observed data and derived, model-free measures for subject 1 following oral administration of 250 mg of amoxicillin (accompanies Fig. 1)*

Subject No. 1

Weight = 97.73 kg

OBSERVED DATA

| Sample Times (min) | Observed Serum Levels ($\mu$g/ml) |
|---|---|
| 20 | <0.5 |
| 40 | 1.7 |
| 60 | 2.5 |
| 90 | 2.8 |
| 120 | 2.5 |
| 240 | 0.5 |
| 360 | <0.5 |

Urinary Recovery = 130.6 mg at 6 h, = 52% of dose

EXPONENTIAL FIT FOR ABSORPTION PHASE

Absorption Delay[a] = 0.33 h
Absorption Rate Constant = 1.57/h, Half Time = 0.44 h

QUADRATIC FIT FOR PEAK

Time to Peak = 1.50 h, Peak = 2.84 $\mu$g/ml

EXPONENTIAL FIT FOR EXCRETION PHASE, Last 3 Points

Excretion Rate Constant = 0.58/h, Half Time = 1.20 h

| | |
|---|---|
| Area Under the Curve[b] (h×$\mu$/ml)–0 to 6 h | = 7.5 |
| >6 h | = 0.4[c] |
| Total | = 7.9 |
| Percent excreted in the Urine–0 to 6 h | = 52.3 |
| >6 h | = 2.6[c] |
| Total | = 54.9 |

[a] Time after administration when absorption process appears to begin.
[b] AUC computed using trapezoid approximation.
[c] Corrections based on extrapolation beyond 6 h using the above computed half-time (1.2 h).

**Least-squares fitting.** Given the above assumptions on model form and the generalized solutions it only remains to find the parameters $K_a$, $K_e$, $K_{12}$, and $K_{21}$ to fit the observed data for each subject. The starting values for these parameters are obtained from the previously described nonparametric analysis. For example, first approximations to alpha and beta were provided, respectively, by the distribution and elimination rate constants as previously defined. In the case of i.v. data, a parameter space gradient technique is used to find A, B, alpha, and beta, such that the difference between observed and model drug levels is minimized. This program iteratively minimizes the sum of the squares of distance between observed drug levels and model solution values until the best fit is achieved (27) (Table 2 and Fig. 5).

Individual plots of this type are produced for each subject and examined at this point. Subjective examination is important in the overall pharmacokinetic analysis. Levy and Hollister have demonstrated that a model which fits averaged data is not necessarily qualitatively or quantitatively best for drug levels from individual subjects (18). We agree and report only parameter averages after kinetic analysis of each subject.

**Statistical analysis.** Using the methods outlined, the serum level for each subject in each trial is described in terms of four or more model parameters. A quantitative estimate of the importance of dosage level, route of administration, and intrasubject variation is obtained through the use of ANOVA (5). In the bioavailability study of i.m. amoxicillin, the principle concern was in the demonstration of equivalent or better availability for the i.m. route; thus, a three-way ANOVA (dose × subject × route) was employed. When a significant effect of dosage on a model parameter has been demonstrated by ANOVA, then this "dose effect" can be described and analyzed by means of regression analysis (9). That is, if the percentage absorbed were found to change significantly as a function of dosage administered, then the least-squares linear regression of $K_a$ on dose or log dose is relevant. Since the reliability, i.e., repeatability of absorption, after i.m. administration was of interest, Bartlet's test for homogeneity of variance was applied to the model parameters (26).

The common use of reporting "significant" results as those achieving $P < 0.05$ or even $P < 0.01$ dates from the precomputer era when tables were necessary to arrive at the $P$ value for a given F ratio. A

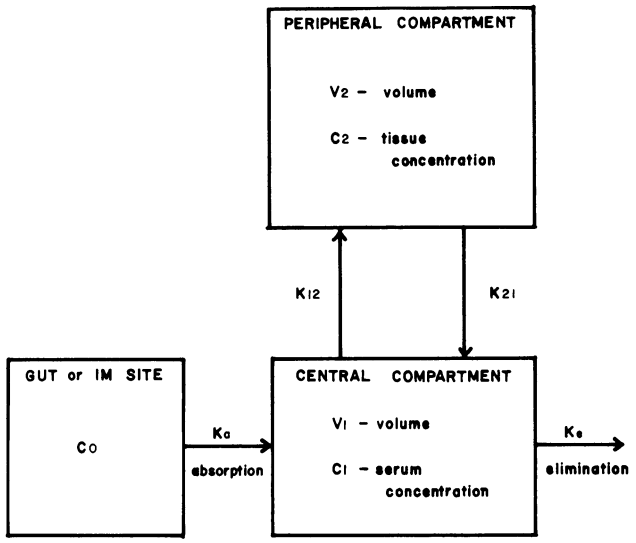


FIG. 2. *Schematic of two-compartment linear pharmacokinetic model. Absorption and elimination are restricted to the central compartment, which is usually considered to represent serum and highly perfused tissue. The peripheral compartment is associated with less well perfused tissue, and thus acts somewhat like a depot for the drug. Each K represents a rate constant of drug transfer and has units of liters per hour. The addition of a third compartment representing the gut (if oral) or injection site (if parenteral) permits description of p.o. or i.m. administration.*

$$C_1(t) = Ae^{-\alpha t} + Be^{-\beta t}$$

Where

$$A = \frac{Do}{V_1} \quad \frac{(K_{21}-\alpha)}{(\beta-\alpha)}$$

$$B = \frac{Do}{V_1} \quad \frac{(K_{21}-\beta)}{(\alpha-\beta)}$$

$$\alpha = \tfrac{1}{2}\,(b + \sqrt{b^2-4K_{21}K_e})$$

$$\beta = \tfrac{1}{2}\,(b - \sqrt{b^2-4K_{21}K_e})$$

$$b = K_{12} + K_{21} + K_e$$

FIG. 3. *Exponential equation for solution for drug level in the central compartment ($C_1$).*

$$C_1(t) = -(P+Q)\,e^{-K_a t} + P\,e^{-\alpha t} + Q\,e^{-\beta t}$$

Where

$$P = \frac{K_a\, f\, Do}{V_1\,(\alpha-\beta)} \quad \frac{(\alpha-K_{21})}{(K_a-\alpha)}$$

$$Q = \frac{K_a\, f\, Do}{V_1\,(\alpha-\beta)} \quad \frac{(K_{21}-\beta)}{(K_a-\beta)}$$

FIG. 4. *First-order equation describing absorption of tablets given p.o.*

few lines of coding in BASIC or FORTRAN are all that is necessary to compute $P$ values (1). In this report, computed $P$ values are used wherever hypothesis testing is involved.

## RESULTS

**Intravenous dosage.** The results of analysis for the drug levels following i.v. administration are summarized in Table 3. This includes the parameter means for each of the five rate constants, clearance in liters per kilogram per hour, the volume distribution for the central compartment, and the total volume distribution. The means are given for the individual 250-, 500-, and 1,000-mg dosages and finally, the overall average across all doses for total trial observations with the standard deviation and SEM are reported. The urinary recovery is reported as percentage of administered dose.

Analysis of variance for this data (Table 3) revealed no significant intrasubject variation, and only clearance exhibited statistically significant dose effect, though neither $K_e$ nor $V_1$ showed significant dose effect (clearance = $K_e \times V_1$).

The averaged parameters ($K_{12}$, $K_{21}$, $K_e$, and $V_1$) were then used in subsequent pharmacokinetic analysis for i.m. and p.o. administration.

TABLE 2. *Sample of two-compartment model analysis. Computer printout of observed levels, predicted levels, and derived model parameters for subject 21 following i.m. administration of 500 mg of amoxicillin (accompanies Fig. 5)*

Subject No. 21

Weight = 81.8 kg

OBSERVED DATA

| Sample Times (min) | Observed Serum Levels ($\mu$g/ml) | Predicted Serum Levels[d] ($\mu$g/ml) |
|---|---|---|
| 20 | 8.09 | 7.72 |
| 40 | 8.80 | 8.61 |
| 60 | 6.91 | 7.25 |
| 90 | 5.71 | 5.18 |
| 120 | 5.18 | 3.67 |
| 240 | 0.97 | 0.93 |
| 360 | <0.50 | 0.24 |

Giving weighted RMS error of 0.64 $\mu$g/ml

MODEL PARAMETERS[b]

Absorption Delay = 0.94 h, Fraction Absorbed = 1.00
Absorption Rate Constant = 2.00/h, Half Time = 0.35 h
Excretion Rate Constant[c] = 1.98/h, K12,[c] K21[c] = 1.77, 1.91/h

MEASURES BASED ON MODEL

Peak Drug Level = 8.81 $\mu$g/ml, Time to Peak = 0.54 h
Area under the curve = 17.3 h×$\mu$g/ml, versus 17.3 for i.v.

[a] Based on model parameters given below.
[b] Chosen to best fit (minimize RMS error) the observed data.
[c] These parameters are based on the i.v. study.

TABLE 3. *Parameters derived using two-compartment model-i.v. administration. Means for each dose and overall means, standard deviations, and SEMs*

| Dose (mg) | Distribution | | | Elimination | | | Volumes | | Urinary Recovery (% dose) |
|---|---|---|---|---|---|---|---|---|---|
| | Alpha (/h) | $K_{12}$ (/h) | $K_{21}$ (/h) | Beta (/h) | $K_e$ (/h) | Clearance (liters/kg/h) | $V_1$ (liters/kg) | $V_d$ (liters/kg) | |
| 250 | 5.90 | 3.05 | 2.76 | 2.55 | 2.65 | 0.32 | 0.17 | 0.29 | 55.8 |
| 500 | 4.04 | 1.43 | 1.64 | 0.72 | 1.68 | 0.32 | 0.22 | 0.46 | 60.0 |
| 1000 | 2.72 | 0.70 | 1.22 | 0.79 | 1.56 | 0.34 | 0.22 | 0.49 | 50.9 |
| Across all doses for 20 trial observations | | | | | | | | | |
| Means | 4.30 | 1.77 | 1.91 | 1.37 | 1.98 | 0.32 | 0.20 | 0.41 | 56.4 |
| Standard deviation | 4.00 | 2.27 | 1.12 | 2.50 | 1.20 | 0.08 | 0.08 | 0.18 | 26.9 |
| SEM | 0.90 | 0.51 | 0.25 | 0.56 | 0.27 | 0.02 | 0.02 | 0.04 | 6.5 |

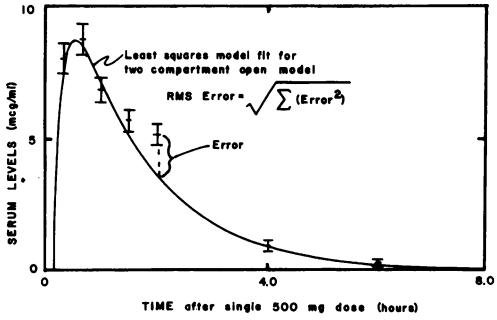


FIG. 5. *Sample plot for two-compartment model fit. Model parameters (rate constants and volumes) are chosen to best match (with minimum RMS error) the observed drug levels. Accompanying printout is contained in Table 2.*

**Bioavailability of i.v. injections.** A separate group of 24 subjects received amoxicillin by i.m. and p.o. routes. Distribution kinetics are assumed to be those determined in the i.v. study (preceding section). Table 4 shows the results of the i.m. administration for each of the three dosages studied, as well as the overall means. Peak and area values are corrected for 70-kg body weight and 1,000-mg dose to facilitate comparison among doses (i.e., data for 250-mg dose are multiplied by 4; data for 500-mg dose are multiplied by 2).

The results of analysis of variance and least-squares regression for i.m. administration, dose effect, subject effect, and regression are included in Table 4. Table 5 presents the equivalent data for p.o. dosage in the same subjects.

TABLE 4. *Analysis of parameters derived using two-compartment model–i.m. administration. Means for each dose, overall means, standard deviations, analysis of variance for dose and subjects, and linear regression for dose*

| Dose (mg) | Absorption | | Peak | | Area | | | Urinary recovery (% dose) |
|---|---|---|---|---|---|---|---|---|
| | Delay (h) | $K_a$ (/h) | Time (h) | Concn (μg/ml) | i.m. (h·μg/ml) | i.v. | i.m./i.v. (%) | |
| 250 | 0.15 | 1.43 | 0.89 | 15.4 | 38.4 | 40.5 | 94.8 | 53.4 |
| 500 | 0.21 | 0.77 | 1.28 | 10.9 | 38.7 | 40.5 | 95.6 | 48.7 |
| 1,000 | 0.21 | 0.76 | 1.33 | 9.7 | 34.5 | 40.5 | 85.2 | 51.6 |
| Across all doses for 72 trial observations | | | | | | | | |
| Mean | 0.19 | 0.98 | 1.16 | 12.0 | 37.1 | 40.5 | 91.7 | 51.3 |
| Standard deviation | 0.10 | 0.69 | 0.51 | 5.5 | 4.3 | 0.0 | 10.5 | 15.4 |
| Two-way analysis of variance–subjects versus dose levels | | | | | | | | |
| Dose[a] | | | | | | | | |
| F = | 4.54 | 14.4 | 12.2 | 14.3 | 13.9 | 0.0 | 13.9 | |
| P = | 0.0160 | 0.00007 | 0.00017 | 0.00007 | .00008 | 1.00 | 0.00008 | |
| Subjects[b] | | | | | | | | |
| F = | 3.25 | 2.40 | 3.74 | 2.87 | 3.26 | 0.00 | 3.26 | |
| P = | 0.00055 | 0.00618 | 0.00017 | 0.00149 | 0.00053 | 1.00 | 0.00053 | |
| Least-squares linear regression (for log dose) | | | | | | | | |
| Intercept[c] | −0.066 | 4.06 | −0.796 | 37.2 | 51.9 | 40.5 | 128 | |
| Slope[c] | 0.097 | −1.14 | 0.731 | −9.35 | −5.43 | 0.000 | −13.4 | |
| F = | 4.06 | 15.4 | 10.8 | 16.0 | 9.61 | 1.85 | 9.61 | |
| P = | 0.0451 | 0.00042 | 0.00197 | 0.00035 | 0.00315 | 0.175 | 0.00315 | |

[a] F ratio and corresponding P value for main effect of dose, 3 and 45 degrees of freedom.
[b] F ratio for main effect of subjects, 24 and 45 degrees of freedom.
[c] Intercept and slope of the regression line for each parameter versus log of dose.

TABLE 5. *Analysis of parameters derived using two-compartment model–p.o. administration. Means for each dose, overall means, standard deviations, analysis of variance for dose and subjects, and linear regression for dose*

| Dose (mg) | Absorption | | Peak | | Area | | | Urinary recovery (% dose) |
|---|---|---|---|---|---|---|---|---|
| | Delay (h) | $K_a$ (/h) | Time (h) | Concn (μg/ml) | p.o. (h·μg/ml) | i.v. | p.o./i.v. (%) | |
| 250 | 0.36 | 1.23 | 1.07 | 14.8 | 39.3 | 40.4 | 97.0 | 49.0 |
| 500 | 0.31 | 0.90 | 1.21 | 11.8 | 37.6 | 40.5 | 93.0 | 49.5 |
| 1,000 | 0.37 | 0.66 | 1.46 | 9.5 | 36.2 | 40.5 | 89.4 | 47.2 |
| Across all doses for 71 trial observations | | | | | | | | |
| Mean | 0.35 | 0.93 | 1.25 | 12.0 | 37.7 | 40.5 | 93.1 | 48.5 |
| Standard deviation | 0.14 | 0.49 | 0.41 | 4.3 | 4.1 | 0.0 | 10.2 | 15.1 |
| Two-way analysis of variance–subjects versus dose levels | | | | | | | | |
| Dose | | | | | | | | |
| F = | 1.11 | 13.8 | 8.5 | 18.9 | 3.38 | 0.00 | 3.37 | |
| P = | 0.338 | 0.00009 | 0.0011 | 0.00001 | 0.0428 | 1.000 | 0.0428 | |
| Subjects | | | | | | | | |
| F = | 0.96 | 2.13 | 2.21 | 2.83 | 1.78 | 0.00 | 1.78 | |
| P = | 0.528 | 0.0154 | 0.0118 | 0.00179 | 0.0495 | 1.000 | 0.0495 | |
| Least-squares linear regression (for log dose) | | | | | | | | |
| Intercept | 0.336 | 3.42 | −0.397 | 34.8 | 49.4 | 40.5 | 122 | |
| Slope | 0.001 | −0.92 | 0.606 | −8.43 | −4.28 | 0.000 | −10.6 | |
| F = | 0.000 | 19.7 | 11.5 | 22.9 | 5.32 | 1.76 | 5.32 | |
| P = | 0.982 | 0.00013 | 0.00154 | 0.00006 | 0.0228 | 0.186 | 0.0228 | |

The statistical comparison of i.m. versus p.o. kinetics is accomplished via three-way ANOVA (subject × dose × route) and Bartlet's test of homogeneity of variance (Table 6).

From these results, it can be seen that the delay is slightly greater ($P = 0.0001$), 0.347 h (p.o.) versus 0.192 h (i.m.), although the time to peak concentration is not statistically different

($P$ = 0.189). The concentration peak is identical, 12.0 μg/ml, for each. The area under the curve is essentially identical ($P$ = 0.296): for the p.o. route, 37.7 h · μg/ml versus 37.1 h · μg/ml for i.m. Examination for homogeneity of variances shows that the standard deviation of delay is larger for p.o. (0.139 versus 0.104 h). For $K_a$ and peak, i.m. shows more spread, but for the time to peak and area there is no statistical difference. Based on the above results, it is concluded that the i.m. preparation provides equivalent bioavailability and, moreover, is more rapidly available.

## DISCUSSION

**Comparison with other intravenous studies.** The kinetics of amoxicillin after i.v. administration of 250 mg over 33-min infusion in two subjects with four replications has been presented by Zarowny et al. (32). They reported distribution constants ($K_{12}$, $K_{21}$, $K_e$) and volumes ($V_1$ and $V_d$) as summarized in Table 7. Within the dosage range studied, a two-compartment linear model does provide a useful description of the distribution and excretion of amoxicillin.

**Comparison with other oral studies.** Previous studies comparing amoxicillin with ampicillin and/or epicillin have shown that amoxicillin gives higher peak serum concentrations after p.o. doses (6, 7, 10, 21, 23). These results are collected in Table 8 and show reasonable agreement, among the seven studies, in peak levels after p.o. administration. AUC is probably the best single bioavailability parameter. This study found a 93% ratio of p.o. to i.v. AUC as compared with 89% by Zarowny (32). Urinary recovery data provides a useful, separate bioavailability parameter. (Urinary recovery is independent of serum levels or pharmacokinetic models.) We found about 50% recovery, compared with other reports of 60%. This may relate to sensitivity of other assay techniques to metabolites that cannot be measured in our method.

TABLE 6. *Three-way analysis of variance (subject × dose × route). Analysis for i.m. versus p.o. and test for homogeneity of variance*

| | Absorption | | Peak | | Area | | |
|---|---|---|---|---|---|---|---|
| | Delay (h) | $K_a$ (/h) | Time (h) | Concn (μg/ml) | i.m. or p.o. (h · μg/ml) | i.v. | Ratio (%) |
| Dose | | | | | | | |
| F = | 0.8 | 23.1 | 14.8 | 25.7 | 13.4 | 0.0 | 13.4 |
| $P$ = | 0.550 | 0.00000 | 0.00002 | 0.00000 | 0.00004 | 1.000 | 0.00004 |
| Subjects | | | | | | | |
| F = | 1.4 | 2.7 | 3.0 | 2.9 | 2.1 | 0.0 | 2.1 |
| $P$ = | 0.145 | 0.00053 | 0.00014 | 0.00020 | 0.00669 | 1.000 | 0.00669 |
| Route | | | | | | | |
| F = | 58.9 | 0.5 | 1.7 | 0.02 | 1.1 | 0.00 | 1.1 |
| $P$ = | 0.00000 | 0.506 | 0.189 | 0.893 | 0.296 | 1.000 | 0.296 |
| Bartlet's test of homogeneity of variance | | | | | | | |
| Route | | | | | | | |
| Chi-square[a] | 5.82 | 7.87 | 2.88 | 4.71 | 0.12 | 0.00 | 0.12 |
| $P$[b] = | 0.0153 | 0.00532 | 0.0861 | 0.0284 | 0.733 | 0.0000 | 0.733 |

[a] Represents a measure of the differences in the observed variation (standard deviation) of each parameter.

[b] $P$ value for the chi-square, 1 degree of freedom. Small value infers a statistical difference in variability of that parameter.

TABLE 7. *Pharmacokinetic studies of amoxicillin–i.v. route*

| Source | Parameters from i.v. administration | | | | | |
|---|---|---|---|---|---|---|
| | $K_{12}$ (/h) | $K_{21}$ (/h) | $K_e$ (/h) | Clearance (liters/kg/h) | $V_1$ (liters/kg) | $V_d$ (liters/kg) |
| Zarowny (32) | 1.29 | 1.94 | 1.43 | 0.245 | 0.187 | 0.282 |
| This study | 1.77 | 1.91 | 1.98 | 0.324 | 0.199 | 0.410 |

TABLE 8. *Pharmacokinetic studies of amoxicillin—p.o. route*

| Source | Parameters from p.o. administration | | | | |
|---|---|---|---|---|---|
| | Dose (mg) | Peak serum level (μg/ml) | AUC | | Urinary recovery (%) |
| | | | (h·μg/ml) | % of i.v. | |
| Neu (21) | 250 | 5.2 | | | 58.0 |
| Zarowny (32) | 250 | 3.5 | 9.2 | 89 | |
| This study | 250 | 3.8 | 9.8 | 97 | 49.0 |
| Neu (21) | 500 | 7.6 | | | 75.2 |
| Croydon (7) | 500 | 10.8 | | | |
| Gorden (10) | 500 | 7.6 | | | 60.0 |
| Philipson (23) | 500 | 6.2 | | | 56.8 |
| This study | 500 | 5.9 | 18.8 | 93 | 49.5 |
| This study | 1,000 | 10.2 | 36.2 | 89 | 47.2 |

**Bioavailability after i.m. administration.** The i.m. administration of several drugs, most notably diphenylhydantoin (25, 31) and benzodiazepines (2, 13), demonstrates unreliable absorption and resulting unpredictable serum levels. The spectrum and clinical efficacy of p.o. amoxicillin (8, 14–17, 19, 20, 22, 28) have been reasonably well established. If amoxicillin is to provide a complete therapeutic alternative to ampicillin, then demonstration of complete and reliable absorption after i.m. administration is important. In this study the mean AUC, delay, peak, and time to peak are compared in Table 9. Both i.m. and p.o. exhibit intrasubject effects in all parameters except delay in p.o. (Tables 4 and 5). Not only were the mean values similar for i.m. versus p.o., but the amount of scatter for the two preparations was similar, as summarized in Table 6.

Probably the best evidence for completeness of absorption after p.o. and i.m. administration is the similarity in urinary recovery. The averages across all trials were 56% for i.v., 51% for i.m., and 49% for p.o. There was no statistical difference between i.v. and i.m., or i.m. and p.o.

The drug administered i.m. was sodium amoxicillin prepared by lyophilization, an early formulation that has been superceded by a preparation that is obtained by precipitation. Though kinetics of the precipitated drug are still being studied, preliminary results suggest that higher peak serum concentrations are reached than with the lyophilized preparation.

**Dose dependence.** A statistically significant dose effect in most parameters for both routes was also apparent. After i.m. injection, the 1,000-mg dose resulted in proportionately lower peak serum levels and AUCs (Table 4). This reduction is expressed by the regression equation. For example, the peak serum level expected is given by the slope and intercept (Table 4), viz.: peak concentration = 37.17 − 9.348·log (dose) (in units of μg/ml per 70 kg per 1,000 mg).

TABLE 9. *Summary comparison of i.m. versus p.o. administration. Values are means ±1.0 standard deviation, averaged across all trials, corrected for 1000-mg dose in 70-kg man*

| Parameter | Route of administration | |
|---|---|---|
| | i.m. | p.o. |
| AUC (h·μg/ml) | 37.1 + 4.2 | 37.7 + 4.1 |
| Delay to absorption (h) | 0.192 + 0.104 | 0.347 + 0.139 |
| Peak serum level (μg/ml) | 12.0 + 5.5 | 12.0 + 4.3 |
| Time to peak (h) | 1.16 + 0.51 | 1.25 + 0.41 |

Very similar results occur in these subjects after p.o. dosage. The peak concentration occurs only slightly later than in i.m. (1.25 versus 1.16 h), and the concentration itself falls from 14.8 μg/ml for the 250-mg p.o. dose to 9.54 μg/ml for the 1,000-mg p.o. dose (Table 4). This is largely explained by the reduction in rate of absorption (1.23/h for 250 mg to 0.66/h for 1,000 mg p.o.). Area ratios (p.o.:i.v.) also are reduced, though proportionately much less, from 95 to 85%. These effects, although statistically significant ($P = 0.00008$) in this study, are probably not clinically significant, since average absorption is still approximately 90% by either route. Figure 6 shows comparison of AUC and delay by dose for i.m. and p.o. administration. As exhibited in Table 6, the only parameter that shows a statistical difference in route is delay, although the time of peak serum levels shows no difference. Figure 7 shows plots of average serum levels for i.v., p.o., and i.m. administration.

Thus, amoxicillin does indeed appear to be rapidly and reliably absorbed after i.m. administration, producing equivalent serum levels, and should prove therapeutically bioequivalent to p.o. amoxicillin or i.m. ampicillin.

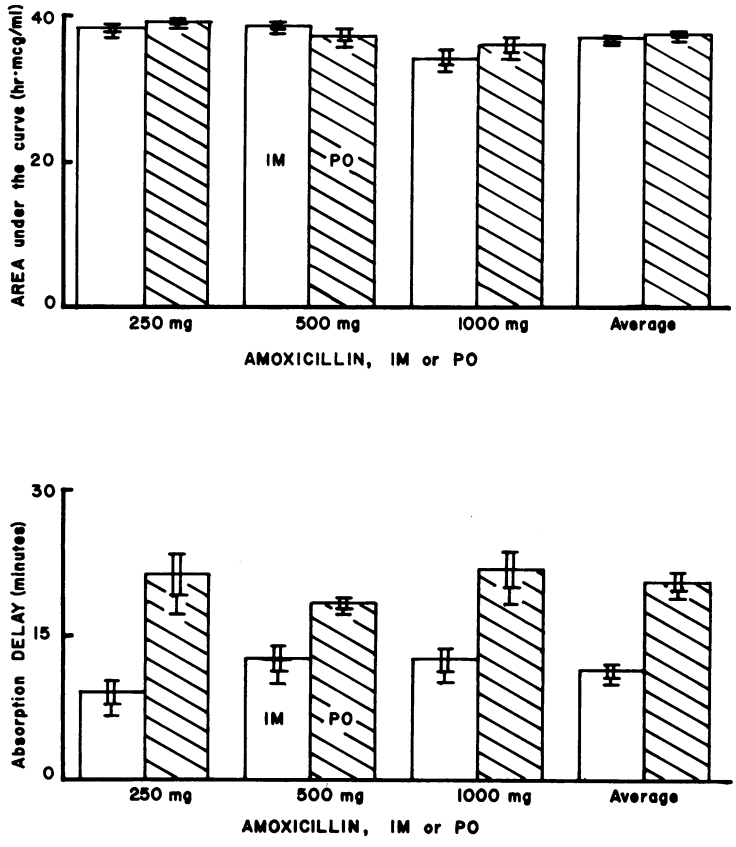


FIG. 6. *Comparison of bioavailability of i.m. and p.o. amoxicillin by dose. Bars indicate the average for all subjects after normalizing to 70-kg body weight and 1,000-mg dose. Double bars (⊡) span ±1.0 SEM for the estimate. Thus, nonoverlapping between two parameters indicates that the parameters are statistically different at the P = 0.05 level. Single bar (I) is −1.96 SEM. If this bar does not reach the base line, then that parameter is statistically nonzero at the P = 0.05 level.*

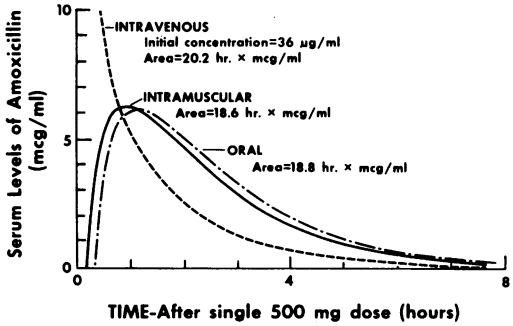


FIG. 7. *Best predictions for serum levels of amoxicillin after i.v., i.m., and p.o. administration. Curves are based on averaged pharmacokinetic parameters assuming a 70-kg patient receiving a single 500-mg dose.*

## ACKNOWLEDGMENTS

We are grateful to Fred Barr, Beecham Laboratories, Bristol, Tenn., for performance of bioassays.

## LITERATURE CITED

1. **Abramowitz, M., and I. A. Stegnum (ed.).** 1970. Handbook of mathematical functions with formulas, graphs, and mathematical tables. U. S. Government Printing Office, Washington, D. C.
2. **Baird, E. S., and D. M. Hailey.** 1972. Plasma levels of diazepam and its major metabolite following intramuscular administration. Br. J. Anaesth. 45:546–548.
3. **Bennett, J. V., J. L. Brodie, E. J. Benner, and W. M. M. Kirby.** 1966. Simplified accurate method for antibiotic assay of clinical specimens. Appl. Microbiol. 14:170–177.
4. **Bischoff, K., and R. L. Dedrick.** 1970. Generalized solution to linear, two-compartment, open model for drug distribution. J. Theor. Biol. 29:63–68.
5. **Bliss, C. I.** 1967. Statistics in biology. McGraw-Hill Book Co., New York.
6. **Bodey, G. P., and J. Nance.** 1972. Amoxicillin: in vitro and pharmacological studies. Antimicrob. Agents Chemother. 1:358–362.
7. **Croydon, E. A., and R. Sutherland.** 1971. α-Amino-p-hydroxybenzyl penicillin (BRL 2333), a new semisynthetic penicillin: absorption and excretion in man, p. 427–433. Antimicrob. Agents Chemother. 1970.
8. **Croydon, E. A. P.** 1973. Clinical experience of amoxy-

cillin in the United Kingdom. Chemotherapy 18(Suppl.):112–118.
9. **Draper, N. R., and H. Smith.** 1966. Applied regression analysis. John Wiley & Sons, New York.
10. **Gordon, R. C., C. Regamey, and W. M. M. Kirby.** 1972. Comparative clinical pharmacology of amoxicillin and ampicillin administered orally. Antimicrob. Agents Chemother. 1:504–507.
11. **Greenblat, D. J., and J. Koch-Weser.** 1975. Clinical pharmacokinetics (second of two parts). N. Engl. J. Med. 293:964–970.
12. **Greenblatt, D. J., and J. Koch-Weser.** 1975. Clinical pharmacokinetics (first of two parts). N. Engl. J. Med. 293:702–705.
13. **Greenblatt, D. J., and R. I. Shader.** 1974. Benzodiazepines (first of two parts). N. Engl. J. Med. 291:1011–1015.
14. **Handsfield, H. H., H. Clark, J. Wallace, K. K. Holmes, and M. Turck.** 1973. Amoxicillin, a new penicillin antibiotic. Antimicrob. Agents Chemother. 3:262–265.
15. **Harding, J. W., and L. J. Less.** 1972. Trial of a new broadspectrum penicillin (amoxycillin) in general practice. Practitioner 209:363–368.
16. **Holloway, W. J.** 1973. Clinical experience with amoxicillin—a preliminary report. Infect. Dis. Rev. 2:245–251.
17. **Leigh, D. A.** 1972. Diagnosis of urinary tract infections in general practice, and treatment with a new penicillin—amoxycillin. Curr. Med. Res. Opin. 1:10–18.
18. **Levy, G., and L. E. Hollister.** 1964. Inter- and intrasubject variations in drug absorption kinetics. J. Pharm. Sci. 53:1392–1403.
19. **Middleton, F. G., D. M. Poretz, and R. J. Duma.** 1973. Clinical and laboratory evaluation of amoxicillin (BRL 2333) in the treatment of urinary tract infections. Antimicrob. Agents Chemother. 4:25–30.
20. **Neu, H. C., and E. B. Winshell.** 1970. In vitro antimicrobial activity of 6[d(−)a-amino-p-hydroxyphenylacetamido] penicillanic acid, a new semisynthetic penicillin, p. 407–410. Antimicrob. Agents Chemother. 1969.
21. **Neu, H. C., and E. B. Winshell.** 1970. Pharmacological studies of 6[D(−)α-amino-*p*-hydroxyphenylacetamido] penicillanic acid in humans, p. 423–426. Antimicrob. Agents. Chemother. 1969.
22. **Neu, H. C.** Just how good is amoxicillin? Med. Times, February 1975.
23. **Philipson, A., L. D. Sabath, and B. Rosner.** 1975. Sequence effect on ampicillin blood levels noted in an amoxicillin, ampicillin, and epicillin triple crossover study. Antimicrob. Agents Chemother. 8:311–320.
24. **Riegelman, S., J. C. K. Loo, and M. Rowland.** 1968. Shortcomings in pharmacokinetic analysis by conceiving the body to exhibit properties of a single compartment. J. Pharm. Sci. 57:117–123.
25. **Serrano, E. E., D. B. Roye, R. H. Hammer, et al.** 1973. Plasma diphenylhydantoin values after oral and intramuscular administration of diphenylhydantoin. Neurology 23:313–317.
26. **Sokal, R. R., and F. J. Rohlf.** 1969. Biometry—the principles and practice of statistics in biological research. W. H. Freeman, San Francisco.
27. **Spyker, D. A.** 1970. Simulation in the analysis and control of a cardio-circulatory assist device. Simulation 15:196–205.
28. **Stam, J.** 1973. Preliminary clinical study with amoxycillin (brl 2333) in complicated lower respiratory tract infections. Chemotherapy 18(Suppl.):27–33.
29. **Sutherland, R., E. A. P. Croydon, and G. N. Rolinson.** 1972. Amoxycillin: a new semi-synthetic penicillin. Br. Med. J. 3:13–16.
30. **Wagner, J. G., and E. Nelson.** 1963. Per cent absorbed time plots derived from blood level and/or urinary excretion data. J. Pharm. Sci. 52:610–611.
31. **Wilensky, A. J., and J. A. Lowden.** 1973. Inadequate serum levels after intramuscular administration of diphenylhydantoin. Neurology 23:318–324.
32. **Zarowny, D., R. Ogilvie, D. Tamblyn, C. Macleod, and J. Reudy.** 1974. Pharmacokinetics of amoxicillin. Clin. Pharmacol. Ther. 16:1045–1051.